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EVALUATION OF THE NITROGEN STATUS OF CORN BY TISSUE TESTS

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EVALUATION OF THE NITROGEN STATUS OF CORN BY TISSUE TESTS

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Tissue testing is recognized as a useful technique for assessing the nutrient status of a growing crop. Tissue testing can detect plant-nutrient deficiencies before visual growth symptoms or reduced yield is noted, thus providing an opportunity for correcting these deficiencies before damage to the crop occurs. The basic principles and methods for various tissue tests were described earlier (3, 4, 5, 6).[3]

Obviously, tissue tests can be of value only if the tests obtained can be interpreted in terms of adequate or inadequate levels of nutrition. This is often the major limitation on their usefulness and this limitation means that before tissue tests can be used successfully, the test procedure employed must be properly correlated with quantitative measurements of the nutrient in question. The research reported in this publication involved the use of a nitrate tissue test on field corn and was carried out to establish correlations between tissue test readings and true N status as determined by N concentration in leaves, total N uptake, and grain yield.

EXPERIMENTAL PROCEDURE

Tissue tests were carried out in conjunction with a N fertility experiment started on the Roza Unit of the Irrigation Experiment Station at Prosser, Wash., in 1955 (1). In the first year 5 rates of N were applied to each of two areas. One area was taken out of native vegetation[4] with no previous cropping, and the other was an adjacent area that had been cropped to alfalfa for 3 years. Since the virgin soil was low in available N, corn growing under 10 levels of N nutrition was available for testing. In 1956 each main plot was subdivided for 6 additional N rates, thereby providing a possible total of 60 N levels for testing in 1956. Treatments were arranged in randomized blocks and replicated 3 times.

The soil type in both areas was Warden very fine sandy loam, a neutral soil that is noncalcareous to approximately 12 inches and underlain with bedrock at 36 to 60 inches. Total N in the virgin soil was low--about 0.041 percent in the plow layer--and the C:N ratio was 9:1. Total N in the alfalfa area plow layer was 0.056 percent.

All tests were made on hybrid field corn Wis. 641-AA, grown uniformly on both areas. Various measures in the form of soil tests or supplemental fertilization were carried out to insure that N would be the only limiting nutrient. Row spacing was 38 inches with plants spaced

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[1] Contribution from the Soil and Water Conservation Research Division, Agricultural Research Service, U.S. Department of Agriculture, in cooperation with the Washington State Agricultural Experiment Station.

[2] Soil Scientist, Research Technician, and formerly Soil Scientist, respectively, USDA, Prosser, Wash. J. L. Nelson is now working in private industry.

[3] Underscored figures in parentheses refer to Literature Cited at end of report.

[4] Primarily bunchgrass, Agropyron inerme.

8 inches in the row, to give a plant population of 17,500 per acre. Irrigation was by furrow, and care was taken to avoid or minimize leaching. Both fields had a uniform slope of about 3 percent.

Tissue tests in 1955 were exploratory and were made at 6 stages of growth, beginning when the plants were approximately 2 feet tall and continuing until 20 days after silking. Also, two different leaves were tested at each growth stage--the second leaf from the ground and the youngest, mature leaf.

Tests made in 1956, which are the basis for correlations given in figures 1 through 3, were made during the period of tasseling on a leaf halfway up the stalk. Four leaves were tested per plot, thus with 3 replications the data reported are averages of 12 individual tissue tests.

The second leaf below the upper ear was sampled for determining leaf N concentration at silking. This sample consisted of 10 leaves per plot. Total N uptake was based on the N content of total tops from two 10-foot rows sampled after the kernels had dented. Yields are for shelled corn at 15.5-percent moisture.

All tissue tests were made with alpha-naphthylamine nitrate test powder prepared according to the directions of Bray (2). A small, uniform quantity of the reagent was placed on the leaf midrib about 3 inches from the stalk. The powder was then wetted with plant sap by crushing the midrib with a pair of pliers. After about 30 seconds the pink color that develops as a reaction product between the nitrates in the plant sap and the test powder was compared with a color card [5] and given an arbitrary numerical rating, with zero indicating no color and nine indicating maximum coloration (7 maximum in 1955).

EXPERIMENTAL RESULTS

Tissue Tests in Relation to Stage of Growth

The results of tissue testing done in 1955 on youngest mature leaves are given in table 1. Tests made on the second leaf from the ground gave similar results. From these data it can be concluded that to obtain a maximum yield (in this case about 160 bu./acre) the tissue test should definitely not indicate a decline in nitrate level prior to tasseling, and that to eliminate any possibility of a yield decrease because of N deficiency, a maximum tissue test should probably be maintained until after silking. Some drop in the nitrate level of the index tissue after silking is not detrimental to yield, as was evidenced by maximum yields from plots that had a low nitrate test 20 days after the silking period.

An additional conclusion supported by the data in table 1 is that a maximum yield of roughly 160 bushels per acre is associated with a leaf N concentration at silking of about 2.9 percent and a total N uptake of approximately 200 pounds per acre. The critical leaf N concentration obtained confirms the values previously reported by Tyner (7) and by Viets and coworkers (8).

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[5] The color card used was part of a tissue test kit manufactured by Urbana Laboratories, Urbana, Ill. Mention of company name is included for the benefit of the reader and does not infer any endorsement or preferential treatment of the product listed by the U.S. Department of Agriculture.

TABLE 1.--Nitrate tissue tests on youngest mature leaves at six stages of growth as related to the N nutrition and yield of corn, 1955.

Area and N rate	Tissue test rating[1] on:						N. Conc. in leaves at silking	Total N uptake	Grain yield
	7/1	7/8	7/15	7/27	8/11	8/31			
Lb./acre Virgin Area:							Pct.	Lb./acre	Bu./acre
0	1.5	0	0	0	0	0	0.89	25.5	20.9
40	4.9	1.6	.2	0	0	0	.95	52.7	55.3
80	7.0	5.8	3.5	.9	0	0	1.54	75.7	86.0
120	7.0	7.0	6.0	3.6	.9	0	1.95	113.0	113.8
200	7.0	7.0	7.0	7.0	5.5	1.8	2.69	166.2	144.2
L.S.D. (at 5-percent levels)							0.21	16.2	19.6
Alfalfa Area:									
0	7.0	7.0	7.0	7.0	5.0	0	2.73	176.7	139.2
40	7.0	7.0	7.0	7.0	5.9	2.6	2.93	204.5	160.5
80	7.0	7.0	7.0	7.0	7.0	5.5	3.10	213.3	147.7
120	7.0	7.0	7.0	7.0	7.0	7.0	3.13	236.7	163.1
200	7.0	7.0	7.0	7.0	7.0	7.0	3.20	240.6	161.0
L.S.D. (at 5-percent level)							0.17	31.8	13.8

[1] Rating for arbitrary units: 0 equals no color or very little nitrates in plant sap; 7 indicates maximum coloration.

Stages of growth: 7/1, corn 2 ft. tall; 7/8, corn 3 ft. tall; 7/15, pre-tasseling; 7/27, tasseling; 8/11, silking; 8/31, post silking.

Correlation of Tissue Test at Tasseling With Leaf N Concentration

Figure 1 shows the relation between 1956 tissue test ratings made at tasseling and the N concentration in leaves at silking. The overall correlation coefficient for this relation is highly significant, indicating that the tissue tests gave an accurate evaluation of the N status. A single correlation as shown is obviously not the most precise evaluation of this data. From a visual examination it is apparent that the relation for the alfalfa area is different from that for the virgin area. Furthermore, it is obvious that the relation in the area of extreme N deficiency (tissue tests from 0 to 1.5) has a much greater slope than the relation at the higher levels of N nutrition. Whether treated on the basis of obviously different groupings or pooled as shown in figure 1, the data establish that a maximum tissue test at tasseling was essential in order to maintain the leaf N concentration at or above the critical level at silking.

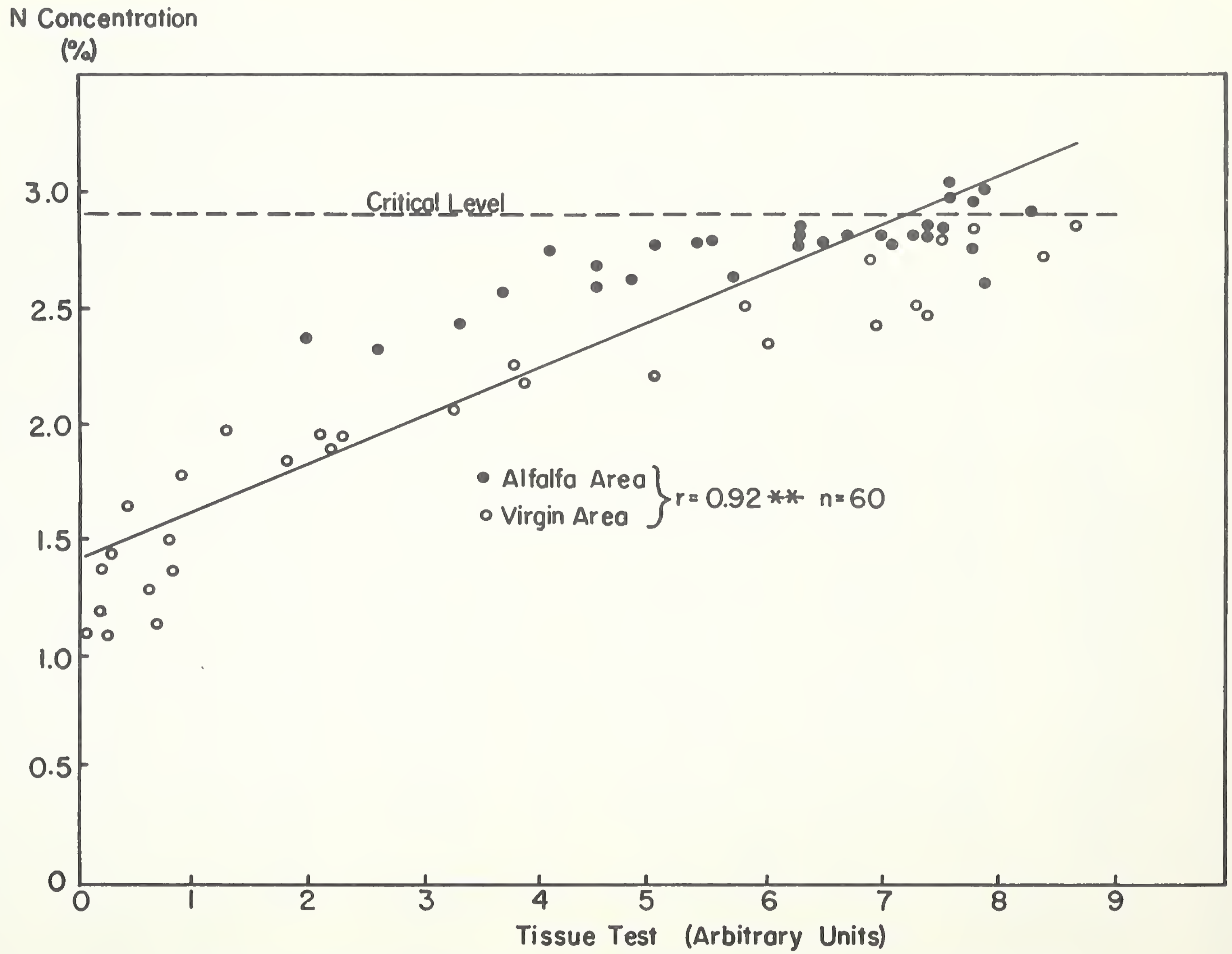

FIGURE 1.--Relation between nitrate tissue tests at tasseling and N concentration in leaves at silking in 1956. The line shown is the calculated regression.

Correlation of Tissue Tests at Tasseling With Total N Uptake

Tissue test correlation with total N uptake is shown in figure 2. This correlation is again highly significant, further substantiating the tissue tests as valid indicators of N status. The relation between tissue tests and N uptake is similar to the relation between tissue tests and N concentration in leaves in that there is a distinct separation between alfalfa area and virgin area data. Also, there is an apparent departure from linearity in the range of extreme N deficiency.

For a given tissue test, total N uptake was roughly 50 pounds greater on the alfalfa area than on the virgin area. Evidently, plants growing on the alfalfa area had higher concentrations of N in tissues not tested or were able to obtain N for a longer period after tasseling than were plants growing on the virgin area. Since the virgin area plants were almost wholly dependent on N from the fertilizer source, their supply of N possibly became exhausted soon after tissue tests were made, whereas it would be reasonable to assume that the large reserves of organic N in the alfalfa area would continue to release nitrate throughout the growing period.

Whatever the reason for the separation of data from the two areas, this observation serves to emphasize that cropping history needs to be considered when interpreting tissue tests, and that correlations obtained under a given N regime may not be strictly valid for another.

Correlation of Tissue Test at Tasseling With Grain Yield

Tissue tests were not nearly so well correlated with grain yields (fig. 3) as with leaf N concentration and total N uptake. Here again, the data from the two areas separate, and considerably larger yields were obtained on the alfalfa area for a given tissue test rating than on the virgin area. On the alfalfa area, yields of 130 bushels of corn per acre were obtained even though the nitrate tissue test at tasseling was below 3. Similar tissue tests on the virgin area are associated with yields of about 80 bushels per acre. These marked differences in yield may have resulted from nutritional and non-nutritional factors other than N associated with the growing of alfalfa. The differences in N status apparent in the data of figures 1 and 2, however, would seem to indicate that the N-supplying patterns of the two areas were quite different, and accounted to a large extent for the discrepancy in yields.

Tissue Test Variation

The data in table 2 are presented to show the extent of plant-to-plant variation in tissue test readings, even within rather small experimental units. Considerable variation is apparent, especially in plots where a N stress is beginning to occur. For instance, where 200 pounds of N per acre was applied in 1956 and the average leaf N concentration was 2.50, tissue test ratings ranged from 2 to 8. These data serve to emphasize that tissue tests need to be made on a considerable number of leaves before a reliable estimate of the general N status of the crop can be obtained.

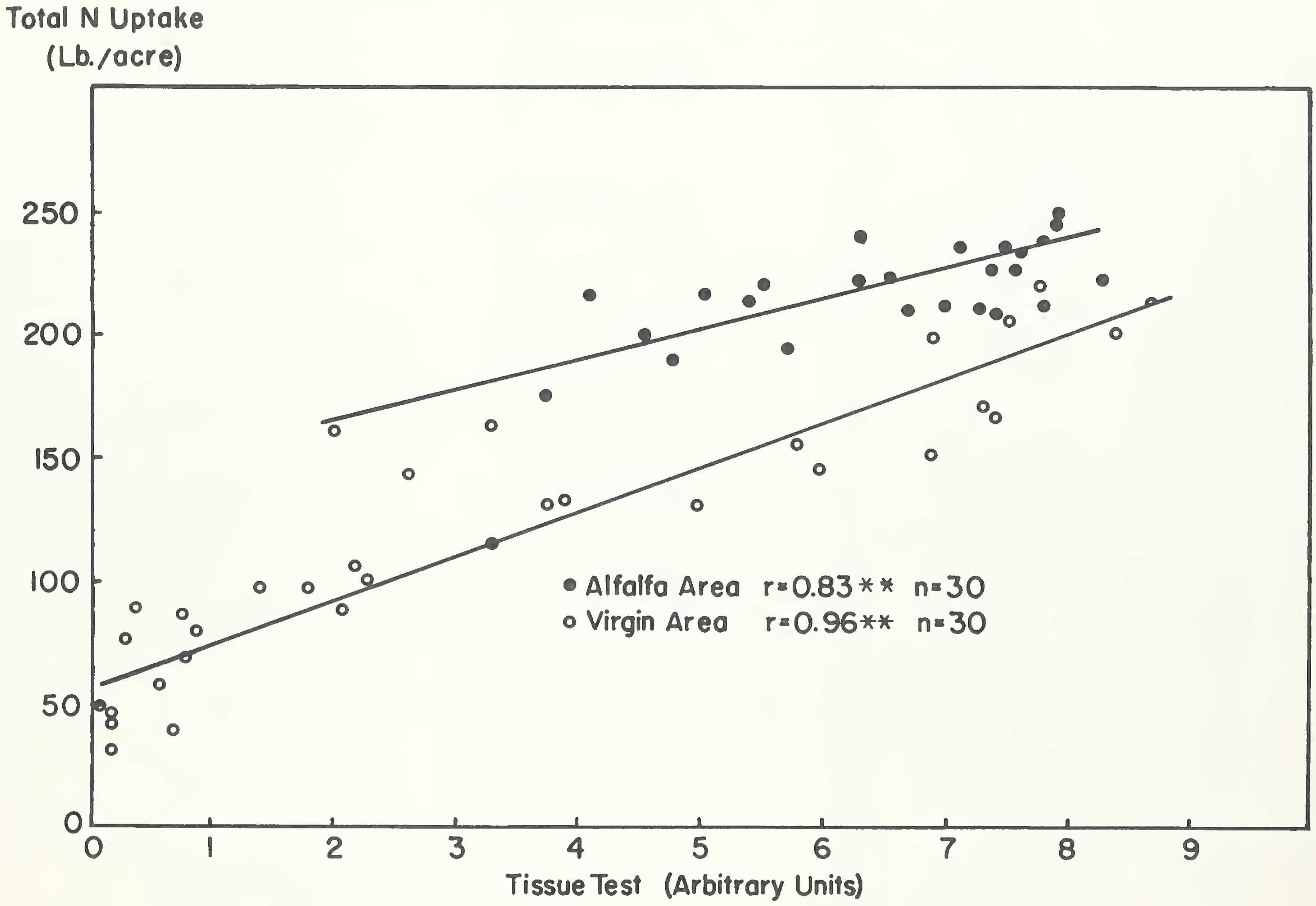

FIGURE 2.--Relation between tissue tests at tasseling and total N uptake in 1956. The lines shown are the calculated regressions.

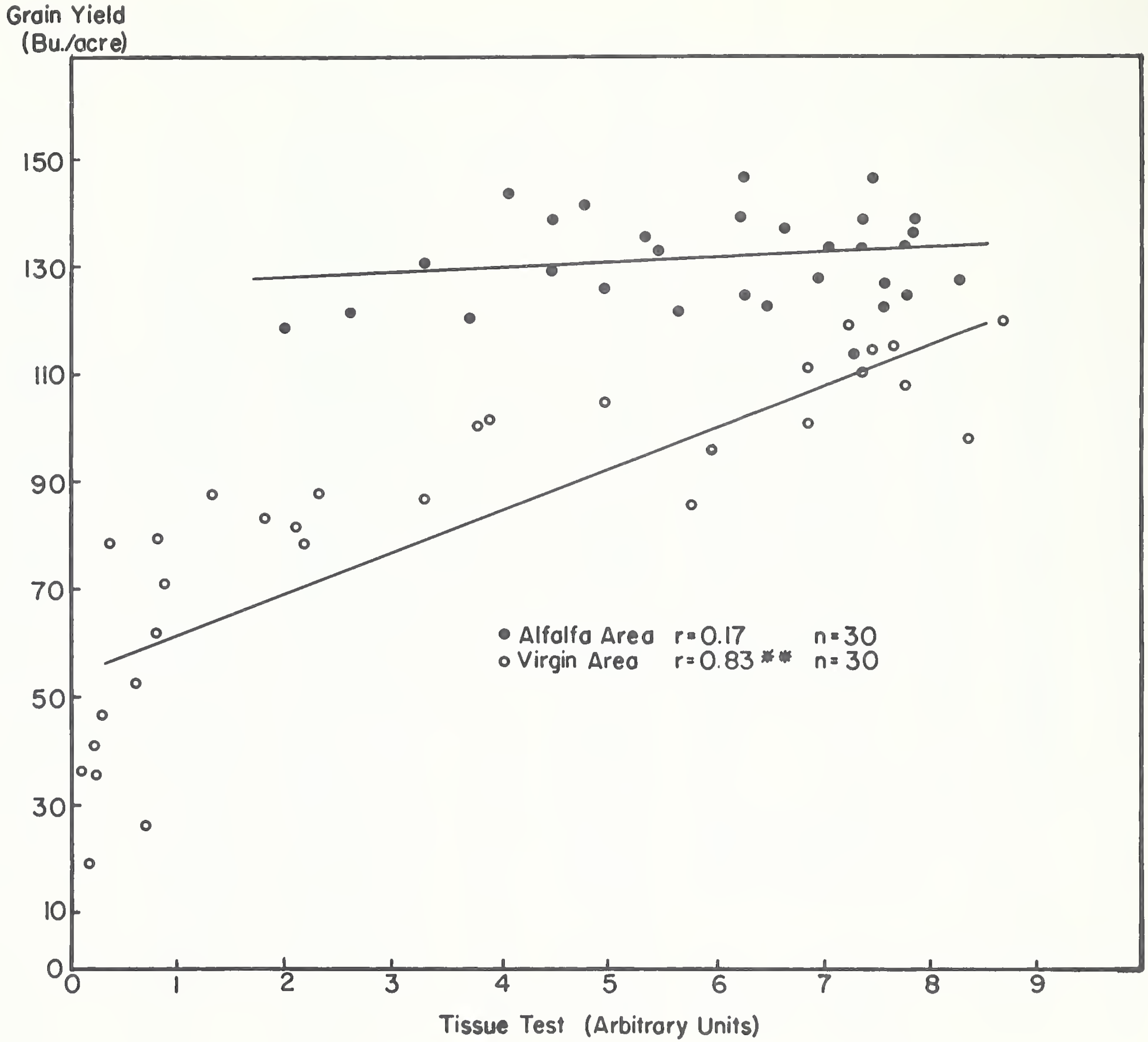

FIGURE 3.--Relation between nitrate tissue tests at tasseling and grain yield in 1956. The lines shown are the calculated regressions.

TABLE 2.--Typical tissue test ratings (arbitrary units from 0 to 9) selected to show the within-treatment variation. (Four tissue tests per plot.)

1955 N rate	1956 N rate	Arbitrary units[1] from replications--			N conc. in leaves at silking
		1	2	3	
		1 2 3 4	1 2 3 4	1 2 3 4	
Lb./acre	Lb./acre				Pct.
80	0	1 0 1 0	0 0 0 0	0 0 0 0	1.19
80	40	1 0 0 1	0 0 0 0	0 1 0 0	1.44
80	80	2 1 1 0	1 1 1 2	0 0 0 0	1.55
80	120	1 7 3 2	1 3 2 3	3 2 0 0	1.90
80	200	6 8 8 7	8 3 5 7	7 2 2 8	2.50
80	400	9 9 7 9	8 9 8 9	8 7 9 9	2.73

[1] Rating for arbitrary units: 0 equals no color or very little nitrates in plant sap; 9 indicates maximum coloration.

SUMMARY

Nitrogen tissue tests were made on field corn growing on plots having possibly 60 different N levels. These tests were correlated with leaf N concentration at silking, total N uptake, and grain yield. The tests were made on the midrib of a leaf halfway up the stalk at tasseling, with alpha-naphthylamine as a reagent.

A highly significant positive correlation ($\underline{r} = 0.92$) was obtained between the tissue tests and leaf N concentration in the second leaf below the upper ear at silking. A maximum tissue test at tasseling was necessary in order to have a leaf N concentration at silking of at least 2.90 percent, which has been shown to be the "critical level" for this nutrient.

A highly significant positive correlation was also obtained between tissue tests and total N uptake. Grain yields on previously uncropped ground, initially low in soil N, were positively correlated with the tissue tests, but on ground where a 3-year-old stand of alfalfa had been plowed down, high yields were obtained even though the tissue test was low at tasseling.

A distinct separation of the correlation data obtained from plots located on virgin ground and from plots located on old alfalfa ground indicated a difference in the N-supplying pattern of the two areas.

Plant-to-plant variation in tissue test ratings was found to be as great as fourfold under conditions of moderate or incipient N deficiency.

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